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Semin Surg Oncol. 1991 Jul-Aug;7(4):221-9.

Biotherapy. 1996;9(1-3):117-21.

Biotherapy. 1996;9(1-3):7-11.

Prog Drug Res. 1994;42:401-21.

Journal of Clinical Hematology and Oncology, (1978) Vol. 8, No. 4, pp.120.

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Cancer Immunology Immunotherapy, (1981) 11/1 (73-79).

Scandinavian journal of immunology, (1974) 3 (2) 223-8.

Christopher Yaen
US Patent Office
Art Unit 1642
571-272-0838
REM 3A20
REM 3C18

13652402

Transfer factor as an adjuvant to non-small cell lung cancer (NSCLC) therapy.

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The rationale for using transfer factor (TF) in lung cancer patients is that the possibility of improving their cell-mediated immunity to tumour associated antigens (TAA) may improve their survival. From Jan 1984 to Jan 1995, 99 non-small cell lung cancer (NSCLC) resected patients were monthly treated with TF, extracted from the lymphocytes of blood bank donors. In the same period, 257 NSCLC resected patients were considered as non-treated controls. The survival rates of the TF treated group appear significantly improved both for patients in stages 3a and 3b, and patients with histological subtype "large cell carcinoma" ($P < 0.02$). Survival of TF treated patients is also significantly higher ($P < 0.02$) for patients with lymph node involvement (N2 disease). The results of this study suggest that the administration of TF to NSCLC resected patients may improve survival.

Publication Types:
- Clinical Trial
- Controlled Clinical Trial

PMID: 8993769 [PubMed - indexed for MEDLINE]

Biotherapy 9: 117–121, 1996.

Transfer Factor as an adjuvant to non-small cell lung cancer (NSCLC) therapy

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Key words: Cell mediated immunity, lung cancer, N2 disease, transfer factor.

Abstract

The rationale for using transfer factor (TF) in lung cancer patients is that the possibility of improving their cell-mediated immunity to tumour associated antigens (TAA) may improve their survival. From Jan 1984 to Jan 1995, 99 non-small cell lung cancer (NSCLC) resected patients were monthly treated with TF, extracted from the lymphocytes of blood bank donors. In the same period, 257 NSCLC resected patients were considered as non-treated controls. The survival rates of the TF treated group appear significantly improved both for patients in stages 3a and 3b, and patients with histological subtype "large cell carcinoma" ($P<0.02$). Survival of TF treated patients is also significantly higher ($P<0.02$) for patients with lymphnode involvement (N2 disease). The results of this study suggest that the administration of TF to NSCLC resected patients may improve survival.

Abbreviations: CMI: cell-mediated immunity; NSCLC: non-small cell lung cancer; TAA: tumour-associated antigens; TF: transfer factor.

Introduction

Cell-mediated immunity (CMI) plays an important role in controlling the proliferation of tumour cells. Since transfer factor (TF) is able to increase CMI, it was tempting to plan clinical trials whereby it could be used to increase cancer patients' cellular immune response to their tumour cells. In 1975, Levin et al. [1] were among the first to produce evidence for in vitro and in vivo transfer of reactivity to tumour cells. They extracted transfer factor from osteosarcoma patients whose lymphocytes were showing cytotoxicity against the tumour cells and injected it into other osteosarcoma patients whose lymphocytes became subsequently cytotoxic and able to kill the autologous tumour cells. At that time, we have shown that specific TF, obtained from patients with high levels of CMI to TAA of bladder carcinoma - as assessed by the leucocyte migration inhibition test - was able to transfer to the leucocytes of the recipient, by in vitro incubation or by in vivo injection, the reactivity observed in the TF donor [2–4]. Such observations encouraged tumour immunologists to treat cancer patients with TF in the hope that the increase of their immune response against TAA could interfere with the tumour growth.

The prognosis of patients with NSC lung cancer remains pessimistic, with an overall 5-year survival rate of 14% for patients in advanced stages[5]. Surgical resection of the tumour remains the principal treatment, but its success is closely related to the stage of the disease at the time of surgery. Despite early diagnosis and improvements in surgical techniques and adjuvant radiotherapy and chemotherapy, the 5- year survival rate ranges from 57–75% for patients in stage I-II to 0– 14% for the advanced stage III-IV [6].

The concept of stimulating the patient's immune system against the tumour has been applied to NSC lung cancer using active immunization with lung cancer TAA, whereas TF has already been used in NSCLC with apparently favourable results both in early and in advanced stages of the disease [7–8]. Additional preliminary results were reported by us [9] and others

Table 1. Surgery and patients' stage in the TF group.

Type of surgery	Total	Stage Ia	II	IIIa	IIIb	IV
Wedge resection	4	3	0	1	0	0
Lobectomy	55	22	12	16	3	2
Bi-lobectomy	8	2	2	3	1	0
Sleeve lobectomy	4	2	2	2	0	0
Sleeve Bi-lobectomy	26	0	0	0	0	0
Pneumonectomy	4	6	4	14	4	0
Total	99	33	20	36	8	2

[10]. Thus, we decided to start a longitudinal study of immunoprophylaxis by treating with TF NSCLC patients immediately after the surgical removal of the primary tumour.

Materials and methods

TF Patients

From Jan 1984 to Jan 1995, 99 consecutive patients (who accepted to give their informed consent) were submitted, after surgery, to adjuvant therapy with TF (87 male, age: 49–60; 12 female, age: 62–80). None of the patients received radio- or chemotherapy pre-operatively. All patients underwent surgery and subsequently were assigned a disease stage according to the new international staging system for lung cancer [13]. Surgical techniques and disease stage of treated patients are shown on Table 1. The extent of pulmonary resection was determined during surgery according to the site and extent of the tumour; wedge resections were performed for insufficient pulmonary function. Ilar and mediastinal lymphnode dissection was carried out in all resected patients. Eighty-nine resections were complete (89.9%), 10 incomplete (4 microscopic and 6 macroscopic residual tumour). As regards the histological type, 36 (36.4%) were adenocarcinomas, 3 (3%) squamous adenocarcinomas, 50 (50.5%) epidermoid carcinomas and 10 large-cell carcinomas (10.1%). Post-operative adjuvant radiotherapy was given to all patients found with metastatic lymphnode involvement (N2 disease) or with incomplete surgical resection.

Control patients

During the same period, 257 consecutive NSCLC resected patients (219 male, age 62±74 yr. and 38 female, age 62±04 yr.) were randomly selected as non treated controls (nt). None of the patients received pre-operatively radio- or chemotherapy. The surgical technique and disease stage are shown in Table 2. Two-hundred-forty-one resections were complete (93.8%), 16 incomplete (7 microscopic and 9 macroscopic residual tumours). With reference to histology, 99 were adenocarcinomas (38,5%), 9 squamous adenocarcinomas (3.5%), 134 epidermoid carcinomas (52.1%) and 15 large-cell type adenocarcinomas (5.8%). In the latter subset of patients, adjuvant radiotherapy was given after surgery to patients with N2 disease or with incomplete resection.

Transfer factor

TF, whose specificities were unknown and thereafter called "unspecific", was prepared from pooled lymphocytes obtained from blood donors' buffy coats using standard techniques. Two units ($2x10^8$cell equiv.) of TF were injected monthly into patients, starting one month after the surgical resection of the tumour. The duration of treatment ranged from 18 to 36 months. After the treatment period, the patients entered a clinical follow-up period.

Statistical analysis

Patients and controls were matched for percentage distribution of sex, histology, surgical technique and tumour stage using the Chi-square test. Age was matched by the Student's *t*- test. Survival was analysed using the Kaplan-Meier life tables method, and each survival curve was matched using Wilcoxon's test. P values ≤ 0.05 were considered significant.

Results

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Table 2. Non-TF-treated patients: surgery and stage.

Type of surgery	total	Stage I	II	IIIa	IIIb	IV
Wedge resection	14	6	0	7	2	0
Lobectomy	150	87	8	51	1	3
Bi-lobectomy	20	9	2	6	2	1
Sleeve lobectomy	10	2	1	7	0	0
Sleeve bi-lobectomy	2	1	0	1	0	0
Pneumonectomy	60	15	6	24	14	1
Total	257	120	17	96	19	5

Table 3. Demography and stage of TF treated patients (TF) and non TF treated controls (nt).

	nt 257 cases	TF 99 cases	P*
mean age	62.02 yr.	60.68 yr.	
Male	85.2%	87.9%	NS
Female	14.8%	12.1%	NS
Stage I	46.7%	33.3%	NS
Stage II	6.6%	20.2%	NS
Stage IIIa	37.4%	36.4%	NS
Stage IIIb	7.4%	8.1%	NS
Stage IV	1.9%	2%	NS
Adeno ca.	38.5%	36.4%	NS
Ad-sq.ca.	3.5%	3%	NS
Epid. ca	52.1%	50.5%	NS
Large cell	5.8%	10.1%	NS

* Chi-square test; NS = not significant; ca.= carcinoma; Ad-sq.= Squamous adenocarcinoma; Epid.= Epidermoid.

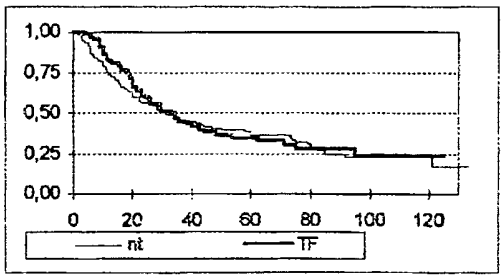

Figure 1. Overall survival of TF patients (TF) and non treated controls (nt).

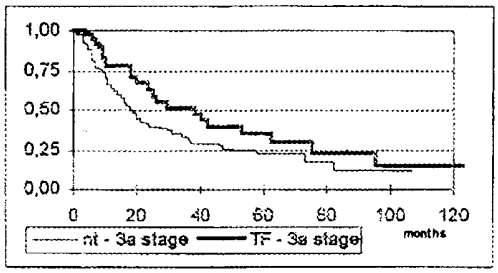

Figure 2. Stage 3a survival rate of TF patients (TF) and non treated controls (nt).

Results

Patients and controls

Mean age and percentage incidence of sex, stage, histology of treated and control patients are shown in Table 3. There were no statistically significant differences between the two groups with respect to age and percentage distribution of sex, stage of disease or tumour histology. (Table 3)

Overall survival

The actuarial survival curves of TF treated control patients do not show significant difference when all stages and histologies are evaluated all together (Fig.1).

Effect of TF according to stage

Significant survival improvement ($P<0.02$) of TF treated patients has been noticed for stage IIIa patients (mean survival 38 months of the TF group, versus 19 of the control group) (Fig.2). In stage IIIb and IV we observed an increased, although not statistically significant, mean survival time in treated patients (IIIb mean survival time: TFT = 24, controls = 15; IV mean survival: TFT = 26, controls = 10).

Effect of TF according to tumour histology

During the evaluation of different histological types (all stages), we observed a significant survival increase of TF treated patients only in the "large cell" carcinoma histological subset ($P<0.02$).

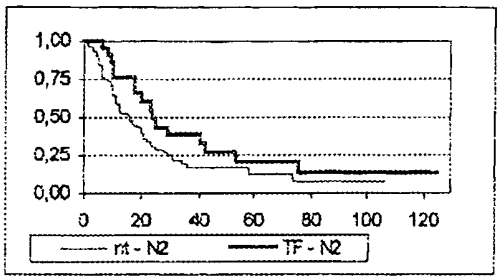

Figure 3. N_2 diseases survival rate of TF patients (TF-N2) and non treated controls (nt-N2).

Effect of TF according to N2 disease

Patients were also evaluated according to the presence of mediastinal lymph node involvement (N2 disease for all tumour stages). The subset of N2 patients receiving TF showed an increased survival ($P<0.02$), compared with the control group (Fig. 3).

Recurrences

The overall recurrence rate was 34.3% in the TF treated patients, and 24.9% in the non-treated controls; the difference is not significant. No difference was found between the two groups when the disease-free interval was considered.

Side effects

None of the patients suffered from any adverse side effect and none discontinued the treatment.

Discussion

The present data confirm observations by others on the efficacy of TF [5,7,8,10,12] and are quite similar to those of Fujisawa and coworkers[12]. These authors treated 263 primary NSCLC cancer patients who had undergone pulmonary resection. They showed that in stage I patients the effect of TF was statistically significant, thus suggesting that TF can suppress "micrometastases" existing at the time of surgery. The authors also primed in vitro lymphocytes from household contacts with IL-2 and mitomycin-treated lung cancer cells. These T-lymphocytes showed considerable cytotoxic activity against the target cells used for in vitro sensitization, and a dialysate obtained from them showed capability to transfer specific cytotoxic activity against lung cancer cells [12].

In another controlled trial in 102 (randomized after surgery) lung cancer patients, the TF group of 44 patients was compared to a control group of 47 patients. Again, the survival of the TF group was significantly better than that of the control in stage I patients, although no significant difference was observed in patients of more advanced stages. Furthermore, significant differences were found between the TF and control groups in patients who had undergone curative resection. The authors concluded that TF seems to inhibit post-operative recurrences and appears to be an effective post-operative adjuvant immunotherapeutic tool for primary resected adenocarcinoma of the lung patients, especially at the early stages [11].

In contrast to the work cited above, Kirsh and coworkers observed good clinical results also in advanced stages patients. In 28 patients with lung cancer, treated at 3-month intervals with 1 ml of TF extracted from the blood of healthy individuals, they showed a significant increase of survival, when the TF-treated group was compared to 35 randomized control patients [7].

The results reported here appear encouraging. Adjuvant immunotherapy using 'unspecific' TF seems to significantly improve the mean survival rate of NSCLC patients in advanced stages whose primary tumour has been removed. The treatment increases the survival of advanced stage patients. It is carried out in out-patients, thus it is low-cost and it does not interfere with the patients' quality of life.

The absence of survival difference in stage I patients is probably due to the long survival of the patients in this stage. Thus, these patients must be monitored for a longer period for a possible difference to appear. Our study used an 'unspecific' TF and a large number of randomized patients. The control group was large enough to offer an accurate picture of the evolution of the disease, when conventional therapies, in the same institution, were applied.

Several questions remain unanswered. For instance, we used a TF from pooled mononuclear cells of blood donors, whose activity may be a mere non-specific stimulation of the patients' CMI. The possibility of an in vitro produced transfer factor, specific for NSCLC antigens, to further improve the survival rates should be investigated. These studies are now in progress.

Acknowledgments

The work was partially funded by the Fondation Asclepios.

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